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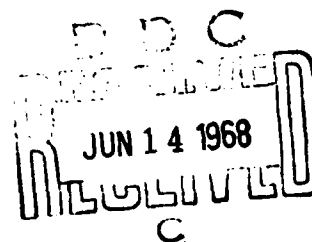
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NEW DATA ON AGGLUTINATION OF ERYTHROCYTES

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# NEW DATA ON AGGLUTINATION OF ERYTHROCYTES\*

Centralblatt für Bakteriologie und Parasitenkunde (Central Journal for Bacteriology and Parasitology), XLVI (1): 49-51, 1908

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In connection with experiments performed jointly with Friedberger to accelerate hemolysis by precipitating sera (Centralblatt für Bakteriologie und Parasitenkunde, Vol XLV, No 4), I performed similar experiments on agglutination. It turned out that the erythrocytes loaded with a dose of the corresponding and nonself-agglutinating immune serum clump together very rapidly and tenaciously if one uses as antiprotein serum a precipitating serum for the animal species which the amboceptor has produced.

In my experiments I used rabbit erythrocytes loaded with amboceptor of a goat treated for some time with rabbit erythrocytes. The serum of a rabbit treated with normal goat serum served as antiprotein serum.

Table 1

(1) Kaninchenblut- körperchen 5 Proz.	(2) Ziegenimmun- serum	(3) Kaninchen- präzipit. Serum	(4) Agglutination
1 cem	0.005 cem	0.0001 cem	(5) 0
1 "	0.005 "	0.0005 "	(5) mäßig
1 "	0.005 "	0.001 "	(6) stark
1 "	0.005 "	0.005 "	(7) sehr stark
1 "	0.005 "	0.01 "	" "
1 "	0.005 "	0.05 "	" "
1 "	0.005 "	0.1 "	" "
1 "	0.01 "	0.1 "	0
(8) 2 Stunden Zimmertemperatur. Zentrifugierung u. Waschung der Blutkörperchen mit Kochsalzlösung (0.85 Proz.)		(9) 2 Stunden Zimmertemperatur	0

1 - Rabbit erythrocytes 5%; 2 - Immune goat serum; 3 - Precipit. rabbit serum; 4 - Agglutination; 5 - moderate; 6 - strong; 7 - very strong; 8 - 2 hours at room temperature. Centrifugation and washing of the erythrocytes with saline solution (0.85%); 9 - 2 hours at room temperature

It will be noted that neither the antiprotein serum by itself regardless of the dose nor the amboceptor serum in the doses under study and multiples thereof is able to agglutinate rabbit red corpuscles.

\* Read on 29 September 1907 at the Berlin Congress for Hygiene and Demography.

In order to enable this special agglutination to show up clearly, it is absolutely essential to work with thoroughly washed, loaded erythrocytes since the presence of all the amboceptor serum, however small the quantity, interferes with the phenomenon. Agglutination occurs only when immune goat serum is used. It is absent when normal goat serum is used, even tho the conditions for precipitation are absolutely the same in both cases (Table II).

Table II

(1) Erythrocyten ke p. 5 Proz.	(2) Zugabe von Serum mit Zugabe normal- serum	(3) Zugabe von präzip. Serum	(4) Agglutination mit (a) Immun- serum (b) Normal- serum
0.001 cem	0.001 cem	0.001 cem	0 0
" 0.005 "	" 0.005 "	" 0.005 "	(5) mäßig 0
" 0.01 "	" 0.01 "	" 0.01 "	(6) stark 0
" 0.05 "	" 0.05 "	" 0.05 "	(7) sehr stark 0
" 0.1 "	" 0.1 "	" 0.1 "	" " 0
" 0.5 "	" 0.5 "	" 0.5 "	" " 0
" 1.0 "	" 1.0 "	" 1.0 "	" " 0
" 0.01 "	" 0.01 "	" 0.01 "	0 0
" 0.05 "	" 0.05 "	" 0.05 "	0 0
" 0.1 "	" 0.1 "	" 0.1 "	0 0
" 0.5 "	" 0.5 "	" 0.5 "	0 0
" 1.0 "	" 1.0 "	" 1.0 "	0 0

(8) 15 min. Zimmertemperatur, Zentrifugung u. Waschung d. Blutkörperchen mit Kochsalzlösung (0.85 Proz.)

(9) 2 Stunden Zimmertemperatur

1-9 - Same as in Table I; 4 - Agglutination with (a) immune serum (b) normal serum

Nevertheless, there is a definite connection. The erythrocytes in contact with the immune serum naturally become loaded with amboceptor, whereas they can draw from the normal serum little or no amboceptor. There still remains the possibility that there is a definitely relationship between the fixed immune amboceptors and the precipitating serum. The following fact indirectly favors this. When heated to 70°, the serum loses both its precipitating ability and its ability to cause the loaded erythrocytes to agglutinate (Table III).

It would be of great value to determine whether precipitation of the precipitin from the antiprotein serum by means of the precipitable substance has removed from the latter the capacity for bringing about agglutination.

According to the experiments performed jointly with Friedberger on accelerating hemolysis, it can also be shown in the case of agglutination that upon contact of the antiprotein serum with the loaded erythrocytes a minimal binding of the substance of importance for agglutination takes place. This substance, however, is difficult to identify and then only after the most careful consideration of the quantitative relations (Table IV).

Table III

(1) Kaninchenblut- körperchen 5 Proz.	(2) Ziegenimmun- serum	(3) Kaninchen- Serum un- oder auf 70° erhitzt	(4) Agglutination (5) un- erhitztes Serum (6) auf 70° er- hitzt
1 ccm	0,005 ccm	0,001 ccm	0
"	0,005 "	0,005 "	0
"	0,005 "	0,01 "	(7) mäßig
"	0,005 "	0,005 "	(8) stark
"	0,005 "	0,1 "	(9) sehr stark
"	0,005 "	0,5 "	" "
"	0,005 "	0,1 "	" 0 "
"	0,01 "	0,1 "	0

(10) 2 Stunden Zimmertemperatur. Zentri-  
fugierung u. Waschung d. Blutkörper-  
chen mit Kochsalzlösung (0,85 Proz.)

(11) 2 Stunden Zimmertemperatur

1 - Rabbit erythrocytes 5%; 2 - Immune goat serum; 3 - Precipit. rabbit serum unheated or heated to 70°; 4 - Agglutination; 5 - unheated serum; 6 - heated to 70°; 7 - moderate; 8 - strong; 9 - very strong; 10 - 2 hours at room temperature. Centrifugation and washing of the erythrocytes with saline solution (0.85%); 11 - 2 hours at room temperature

Table IV

(1) Kaninchenblut- körperchen	(2) Ziegenimmun- serum	(3) Kaninchen präzipit. Serum ausgefällt oder nicht ausgefällt	(4) Agglutination (5) ausgefallenes Serum (6) nicht ausge- fallenes Serum
1 ccm	0,005 ccm	0,001 ccm	0
"	0,005 "	0,002 "	0
"	0,005 "	0,001 "	0
"	0,005 "	0,001 "	(8) Spuren
"	0,005 "	0,002 "	(7) mäßig
"	0,005 "	0,001 "	(9) stark
"	0,005 "	0,006 "	"
"	0,005 "	0,008 "	(10) sehr stark

(11) 2 Stunden Zimmertemperatur. Zentri-  
fugierung, Waschung der Blutkörper-  
chen mit Kochsalzlösung (0,85-proz.)

(12) 0,1 ccm Kaninchen präzipitierendes Serum wurde 2mal mit beladenen (0,01 Ziegen-  
immunserum per Kubikcentimeter) Kaninchenblutkörperchen einer 5-proz. Aufschwemmung  
angefällt und zwar jedesmal mit den Blutkörperchen von 10 ccm dieser Aufschwemmung.

(13) 2 Stunden Zimmertemperatur

1 - Rabbit erythrocytes; 2 - Immune goat serum; 3 - Precipit. rabbit serum precipitated or not precipitated; 4 - Agglutination; 5 - precipitated serum; 6 - nonprecipitated serum; 7 - moderate; 8 - Traces; 9 - strong; 10 - very strong; 11 - 2 hours at room temperature. Centrifugation and washing of the erythrocytes with saline solution (0.85%); 12 - 0.1 cc of precipitating rabbit serum was precipitated twice with loaded (0.01 immune goat serum per cc) rabbit erythrocytes in a 5% suspension and each time with the erythrocytes in 10 cc of this suspension

The facts that I have reported here on rabbit-goat amboceptor, the corresponding erythrocytes and the corresponding rabbit protein serum should also apply to other species of animals.

Likewise the specificity of the reaction should be the same, although further research might disclose whether the phenomenon appears in bacteria the same way that it does in erythrocytes.

It is unjustified, in my opinion, to look upon the data summarized above as a new starting point for explaining or offering theoretical interpretations of the rather delicate processes involved in agglutination.